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(71) Applicant: MERCK & CO., INC. [US/US]; 126 East Lincoln Avenue, Rahway, NJ 07065 (US).

(72) Inventors: AMATO, Joseph, S.; 323 Clinton Street, Brooklyn, NY 11231 (US). CVETOVICH, Raymond; 329 Fawnridge Drive, Scotch Plains, NJ 07076 (US).

(74) Common Representative: MERCK & CO., INC.; Patent Dept., 126 East Lincoln Avenue, Rahway, NJ 07065 (US).

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(54) Title: STABLE SOLVATES OF AVERMECTIN COMPOUNDS

(57) Abstract

There is disclosed a novel form of avermectin compounds wherein the avermectin compounds are crystallized as alcohol solvates to greatly enhance stability of the avermectin drug during long-term storage. The avermectin compounds have utility as highly potent antiparasitic, insecticidal, and anthelmintic agents and compositions for that use are also disclosed.

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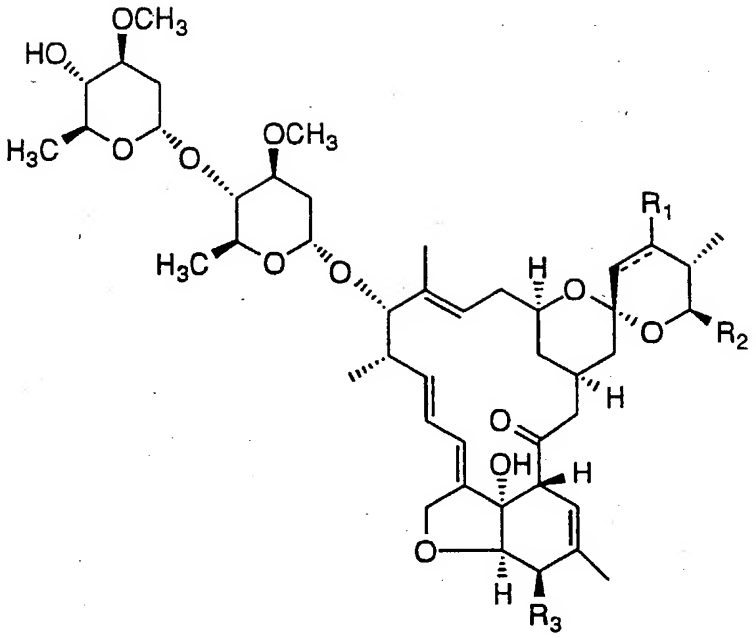
TITLE OF THE INVENTION

STABLE SOLVATES OF AVERMECTIN COMPOUNDS

BACKGROUND OF THE INVENTION

The term avermectin (previously referred to as C-076) is used to describe a series of compounds isolated from the fermentation broth of an avermectin producing strain of Streptomyces avermitilis and derivatives thereof. The morphological characteristics of the culture are completely described in U.S. Patent No. 4,310,519. The natural avermectin compounds are a series of macrolides, each of which is substituted therein at the 13-position with a 4-(α–L-oleandrosyl)-α–L-oleandrose group. The preparation and properties of synthetic avermectin aglycones in which the disaccharide moiety has been removed leaving a free hydroxyl group at position 13 have been described by Mrozik et al., J. Org. Chem. 1982, 47, 489-492 and by Chabala et al., J. Med. Chem. 1980, 23, 1134-1136. Additionally, U.S. Pat. No. 4,199,569 reveals the 22,23-dihydro avermectin compounds. The avermectin compounds and the instant derivatives thereof have a very high degree of anthelmintic and anti-parasitic activity. The natural compounds have the following general structure:



wherein the broken line at the 22,23-position indicates a single or double bond and;

R_1 is	hydroxy and is present only when said broken line indicates a single bond;
R_2 is	isopropyl or sec-butyl; and
R_3 is	methoxy or hydroxy.

There are eight major natural avermectin compounds, designated A1a, A1b, A2a, A2b, B1a, B1b, B2a and B2b. These designations are based on the structure of the individual compounds as shown in the following table (referring to the foregoing structural formula).

Compound	22,23-bond	R_1	R_2	R_3
A1a	double bond	---	sec-butyl	$-OCH_3$
A1b	double bond	---	isopropyl	$-OCH_3$
A2a	single bond	-OH	sec-butyl	$-OCH_3$

A2b	single bond	-OH	isopropyl	$-OCH_3$
B1a	double bond	---	sec-butyl	-OH
B1b	double bond	---	isopropyl	-OH
B2a	single bond	-OH	sec-butyl	-OH
B2b	single bond	-OH	isopropyl	-OH

The avermectins are generally isolated as mixtures of the a and b components (typically $\geq$80% a and $\leq$20% b). Such compounds differ only in the nature of the R_2 substituent and this minor structural difference has been found to have very little effect on the chemical reactivity or biological activity of the compounds. Thus although the a and b components can be separated from each other by chromatography this is not necessary and hence is not normally done. The presence of a mixture of a and b components may be indicated by dropping the a or b from the designation of the compound. A mixture of avermectin B1a and avermectin B1b is thus referred to as avermectin B1. Alternatively a slash(/) is inserted between the compound designations to indicate a mixture such as in "B1a/B1b".

The above structural formula is shown without a definitive stereochemistry at certain positions and with a defined stereochemistry at other positions. However, during the course of the synthetic procedures used to prepare such compounds, or using racemization or epimerization procedures known to those skilled in the art, the products of such procedures can be a mixture of stereoisomers. In particular, the stereoisomers at the 13- and 23-positions may be oriented either α- or β– representing such groups being below or above the general plane of the molecule, respectively. In each such case, and at other positions in the molecule, both the α- and β- configurations are intended to be included within the ambit of this invention.

A related family of natural products is known as the milbemycins. The milbemycins have the same macrocyclic ring structure as the avermectins but have no substitution at position 13 and have a methyl or ethyl group at position 25 (R_2 = methyl or ethyl rather than isopropyl or sec-butyl as in the avermectins). The

milbemycins and the fermentation conditions used to prepare them are described in U.S. Pat. No. 3,950,360. Closely related 13-deoxyavermectin aglycones are prepared by chemical modification of the natural avermectins and have been described in U.S. Pat. Nos. 4,171,134 and 4,173,571.

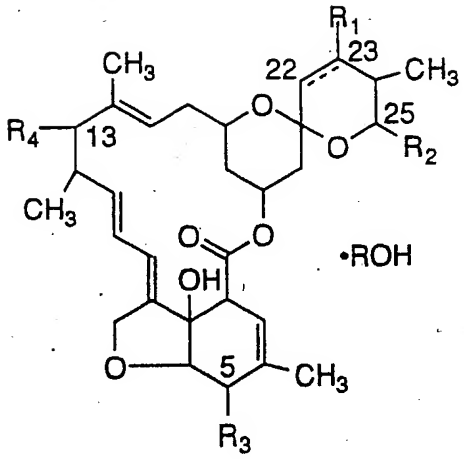
Stabilization of the avermectin class of compounds depends on the particular compound of interest and the method of stabilization. For example, some avermectin compounds require the addition of antioxidants such as propyl gallate, BHA (butylated hydroxy anisole), BHT (butylated hydroxy toluene), monothioglycerol and the like, to the bulk product to inhibit degradation. Other avermectins have been stabilized by the formation of benzoate salts. The present invention is different in that stabilization of avermectins is significantly increased by recrystallization of the product with a sterically encumbered alcohol which results in a new form of avermectin molecule whereby the spatial arrangement of the alcohol in the crystal leads to enhanced thermal stability.

SUMMARY OF THE INVENTION

The instant invention is concerned with a novel form of avermectin compounds wherein the avermectin compounds are recrystallized as alcohol solvates which provides a stable bulk product at ambient temperatures during long-term storage. Thus it is an object of the present invention to describe such stable avermectin alcohol solvates. A further object is to describe processes for the preparation of such compounds. A still further object is to describe the uses of such compounds as anti-parasitic agents and anti-bacterial agents. Still further objects will become apparent from a reading of the following description.

DETAILED DESCRIPTION OF THE INVENTION

The compounds of the instant invention have the following structural formula:



where the broken line indicates a single or a double bond at the 22,23-positions;

R is alkyl of from 1 to 4 carbon atoms resulting in an alcohol consisting of methanol, ethanol isobutanol, isopropanol, propanol or butanol;

R_1 is hydrogen or hydroxy, and is hydroxy only when the broken line indicates a single bond;

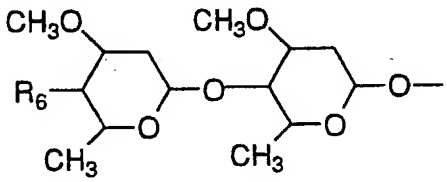
R_2 is alkyl of from 1 to 6 carbon atoms or alkenyl of from 3 to 6 carbon atoms or cycloalkyl of from 3 to 6 carbon atoms;

R_3 is hydroxy, methoxy, or =NOR5;

R5 is hydrogen or methyl; and

R_4 is hydrogen, (C1-C3 alkoxy)(-C0-C3 alkoxy)methoxy,

or



where R_6 is hydroxy, C_1-C_3 amino, or C_2-C_3 alkanoylamino.

The preferred avermectin compounds of the instant invention are the 22,23-dihydro-avermectins and the 13-polyalkoxy avermectin aglycones, most preferably 22,23-dihydro-13-0-[(2-methoxyethoxy)-methyl]avermectin-B1-aglycone (13-O-MEM AVM B1).

The term "loweralkyl" when used in the instant application is intended to represent those alkyl groups either straight or branched chain which have from 1-5 carbon atoms. Examples of such alkyl groups are methyl, ethyl, propyl, iso-propyl, butyl, sec-butyl, pentyl, and the like.

The term "loweralkanoyl" is intended to include those alkanoyl groups containing from one to five carbon atoms in either a straight or branched chain. Examples of such alkanoyl groups are formyl, acetyl, propenyl, butyryl, valeryl, and the like.

The alcohols used in the instant invention are the C_1 thru C_4 alcohols such as ethanol, isobutanol, methanol, isopropanol and the like, most preferrably isopropanol.

PREPARATION OF STARTING MATERIALS

The starting materials for this invention are disclosed in Albers-Schonberg et al., J. Am. Chem. Soc. 1981, 103, 4216-4221 and references cited therein (naturally occurring avermectins), Chabala et al., J. Med. Chem. 1980, 23, 1134-1136 (22,23-dihydro avermectin B1 (Ivermectin), and 22,23-dihydro avermectin B1-aglycone, and US. Patent No. 4,587,247.

The novel compounds of this invention are potent anthelmintic and anti-parasitic agents against internal and external parasitic infestations and are prepared as stable alcohol solvates by the following procedures:

The 22,23 dihydro avermectin aglycone (1 part) is dissolved in isopropanol (1-20 parts) under nitrogen at temperatures ranging from 50°C to 75°C, preferably from 60°C to 70°C and stirring the mixture thus formed at this temperature for 10 to 60 minutes. To

the mixture thus generated is added water (0-20 parts). The intended volume of water can be added in part or whole and the amount added at this point is added at a rate such that a temperature above 50°C is maintained. The resultant solution is then cooled to from about 30°C to 40°C, preferably from about 35°C to 40°C over a period from about 1/2 hour to 1 1/2 hours. The solution is then seeded with from about 0.005 grams to 0.1 grams of 22,23 dihydro avermectin aglycone or the isopropanol solvate thereof. After seeding, the mixture is cooled to from about 25°C to 15°C, preferrably 20°C, over a period of 3 to 20 hours and aged at this temperature from about 0 to 24 hours. To the cooled seeded solution, if the intended volume of water is added in part, is added the final part (volume) of water over a period of 15 to 30 minutes. The resulting slurry is then cooled to from about 0 to 5°C and aged at from about 0 to10°C for approximately 0 to 6 hours. The crystals are then filtered and washed in a 1:1 isopropanol/water solution and dried at room temperature under inert conditions.

The instant compounds of this invention are unexpectedly potent antiparasitic agents against endo and ecto parasites, particularly helminths and arthropods, which cause numerous parasitic diseases in humans, animals and plants.

Parasitic diseases may be caused by either endoparasites or ectoparasites. Endoparasites are those parasites which live inside the body of the host, either within an organ (such as the stomach, lungs, heart, intestines, etc.) or simply under the skin. Ectoparasites are those parasites which live on the outer surface of the host but still draw nutrients from the host.

The endoparasitic diseases generally referred to as helminthiasis are due to infection of the host with parasitic worms known as helminths. Helminthiasis is a prevelant and serious worldwide economic problem due to infection of domesticated animals such as swine, sheep, horses, cattle, goats, dogs cats, and poultry. Many of these infections are caused by the group of worms described as nematodes which cause diseases in various species of animals throughout the world. These diseases are frequently serious and can result in the

death of the infected animal. The most common genera of nematodes infecting the animals referred to above are Haemonchus, Trichostrongylus, Ostertagia, Nematodirus, Cooperia, Ascaris, Bunostomum, Oesophagostomum, Chabertia, Trichuris, Strongylus, Trichonema, Dictyocaulus, Capillaria, Heterakis, Toxocara, Ascaridia, Oxyuris, Ancylostoma, Uncinaria, Toxascaris, and Parascaris. Many parasites are species specific (infect only one host) and most also have a preferred site of infection within the animal. Thus Haemonchus and Ostertagia primarily infect the stomach while Nematodirus and Cooperia mostly attack the intestines. Other parasites prefer to reside in the heart, eyes, lungs, blood vessels, and the like while still others are subcutaneous parasites. Helminthiasis can lead to weakness, weight loss, anemia, intestinal damage, malnutrition, and damage to other organs. If left untreated these diseases can result in the death of the animal.

Infections by ectoparasitic arthropods such as ticks, mites, lice, stable flies, hornflies, blowflies, fleas, and the like are also a serious problem. Infection by these parasites results in loss of blood, skin lesions, and can interfere with normal eating habits thus causing weight loss. These infections can also result in transmission of serious diseases such as encephalitis, anaplasmosis, swine pox, and the like which can be fatal.

Animals may be infected by several species of parasite at the same time since infection by one parasite may weaken the animal and make it more susceptible to infection by a second species of parasite. Thus a compound with a broad spectrum of activity is particularly advantageous in the treatment of these diseases. The compounds of this invention have unexpectedly high activity against these parasites, and in addition are also active against Dirofilaria in dogs, Nematospiroides and Syphacia in rodents, biting insects, and migrating diperous larvae such as Hypoderma sp. in cattle, and Gastrophilus in horses.

The instant compounds are also useful against endo and ecto parasites which cause parasitic diseases in humans. Examples of such endoparasites which infect man include gastro-intestinal parasites of the

genera Ancylostoma, Necator, Ascaris, Strongyloides, Trichinella, Capillaria, Trichuris, Enterobius, and the like. Other endoparasites which infect man are found in the blood or in other organs. Examples of such parasites are the filarial worms Wucheria, Brugia, Onchocerca, and the like as well as extra-intestinal stages of the intestinal worms Strongylides and Trichinella. Ectoparasites which parasitize man include arthropods such as ticks, fleas, mites, lice, and the like and, as with domestic animals, infections by these parasites can result in transmission of serious and even fatal diseases. The instant compounds are active against these endo and ecto parasites and in addition are also active against biting insects and other dipterous pests which annoy humans.

The instant compounds are also useful against common household pests such as Blatella sp. (cockroach), Tineola sp. (clothes moth), Attagenus sp. (carpet beetle), Musca domestica (housefly) and against Solenopsis Invicta (imported fire ant).

The compounds are furthermore useful against agricultural pests such as aphids (Acyrthiosiphon sp.), locusts, and boll weevils as well as against insect pests which attack stored grains such as Tribolium sp. and against immature stages of insects living on plant tissue. The compounds are also useful as a nematodicide for the control of soil nematodes which may be agriculturally important.

For use as an antiparasitic agent in animals the instant compounds may be administered internally either orally or by injection, or topically as a liquid drench or as a shampoo.

For oral administration, the compounds may be administered in capsule, tablet, or bolus form or alternatively they can be mixed in the animals feed. The capsules, tablets, and boluses are comprised of the active ingredient in combination with an appropriate carrier vehicle such as starch, talc, magnesium stearate, or di-calcium phosphate. These unit dosage forms are prepared by intimately mixing the active ingredient with suitable finely-powdered inert ingredients including diluents, fillers, disintegrating agents, and/or binders such that a uniform mixture is obtained. An inert ingredient is one that will not

react with the instant compounds and which is non-toxic to the animal being treated. Suitable inert ingredients include starch, lactose, talc, magnesium stearate, vegetable gums and oils, and the like. These formulations may contain a widely variable amount of the active and inactive ingredients depending on numerous factors such as the size and type of the animal species to be treated and the type and severity of the infection. The active ingredient may also be administered as an additive to the feed by simply mixing the compound with the feedstuff or by applying the compound to the surface of the feed. Alternatively the active ingredient may be mixed with an inert carrier and the resulting composition may then either by mixed with the feed or fed directly to the animal. Suitable inert carriers include corn meal, citrus meal, fermentation residues, soya grits, dried grains and the like. The active ingredients are intimately mixed with these inert carriers by grinding, stirring, milling, or tumbling such that the final composition contains from 0.001 to 5% by weight of the active ingredient.

The compounds may alternatively be administered parenterally via injection of a formulation consisting of the active ingredient dissolved in an inert liquid carrier. Injection may be either intramuscular, intraruminal, intratracheal, or subcutaneous. The injectable formulation consists of the active ingredient mixed with an appropriate inert liquid carrier. Acceptable liquid carriers include the vegetable oils such as peanut oil, cotton seed oil, sesame oil and the like as well as organic solvents such as solketal, glycerol formal and the like. As an alternative, aqueous parenteral formulations may also be used. The vegetable oils are the preferred liquid carriers. The formulations are prepared by dissolving or suspending the active ingredient in the liquid carrier such that the final formulation contains from 0.005 to 10% by weight of the active ingredient.

Topical application of the instant compounds is possible through the use of a liquid drench or a shampoo containing the instant compounds as an aqueous solution or suspension. These formulations generally contain a suspending agent such as bentonite and normally will also contain an antifoaming agent. Formulations containing from 0.005

to 10% by weight of the active ingredient are acceptable. Preferred formulations are those containing from 0.01 to 5% by weight of the instant compounds.

The instant compounds are primarily useful as antiparasitic agents for the treatment and/or prevention of helminthiasis in domestic animals such as cattle, sheep, horses, dogs, cats, goats, swine, and poultry. They are also useful in the prevention and treatment of parasitic infections of these animals by ectoparasites such as ticks, mites, lice, fleas and the like. They are also effective in the treatment of parasitic infections of humans. In treating such infections the compounds of these invention may be used individually or in combination with each other or with other unrelated antiparasitic agents. The dosage of the instant compounds required for best results depends on several factors such as the species and size of the animal, the type and severity of the infections, the method of administration and the compound used. Oral administration of the instant compounds at a dose level of from 0.0005 to 10 mg per kg of animal body weight, either in a single dose or in several doses spaced a few days apart, generally gives good results. A single dose of one of the instant compounds normally gives excellent control however repeat doses may be given to combat re-infection or for parasite species which are unusually persistent. The techniques for administering these compounds to animals are known to those skilled in the veterinary field.

The compounds of these invention may also be used to combat agricultural pests which attack crops either in the field or in storage. The compounds are applied for such uses as sprays, dusts, emulsions and the like either to the growing plants or the harvested crops. The techniques for applying these compounds in this manner are known to those skilled in the agricultural arts.

The following example is provided in order that this invention might be more fully understood; it is not to be construed as limitative of the invention. The avermectin derivative prepared in the following example is characterized using techniques such as High

Performance Liquid Chromatography (HPLC), X-ray crystallography, and the like.

EXAMPLE 1

22,23-Dihydro-13-O-[(2-methoxyethoxy)methyl) Avermectin-B1-Aglycone (13-O-MEM AVM B1) Isopropanol Solvate

50 grams of 13-O-MEM AVM B1 was dissolved in 500 ml of isopropanol under a N_2 stream. The slurry was heated to 65°C to obtain a clear yellow solution. To this solution was added 350 ml of water at a rate to keep the temperature of the solution above 50°C. The solution was allowed to cool to 39°C and then seeded with 50 milligrams of 13-O-MEM AVM B1 isopropanol solvate. The seeded solution was then cooled from 39°C to 30°C at a rate of 3°C per half hour and from 30°C to 21°C at a rate of 1°C per hour. The cooled seeded solution was then contacted with the final volume of water (150 mls) which was added over a 20 minute period and resulted in a slurry of 13-O-MEM AVM B1 crystals. The slurry was cooled to 0 ± 5°C and aged at 0 ± 10°C for three hours. The crystals were filtered and washed with 2 x 50 mls of isopropanol/H_2O (1:1) and dried using a nitrogen stream to yield 58.5 grams of stable 13-O-MEM AVM B1 isopropanol solvate. The HPLC area % and weight % were 97.3% and 97%, respectively.

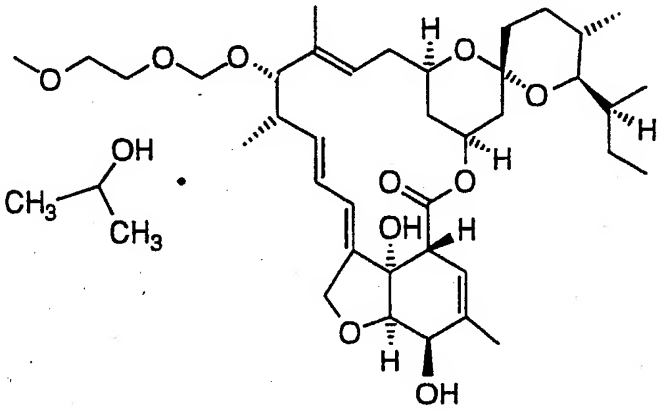
EXAMPLE 2

13-O-MEM AVM B1 Isopropanol Solvate

25 grams of 13-O-MEM AVM B1 was added to 300 ml of isopropanol and heated to 65°C under nitrogen to obtain a clear solution. Water (200 ml) was added keeping the temperature above 50°C. The solution was cooled to 39°C over 30 minutes then seeded with crystalline 13-O-MEM AVM B1 isopropanol solvate (5 mg). The temperature of the mixture was cooled to 20°C over 3 hours and aged at 20°C for 14 hours and the slurry was cooled to 5°C and aged at 5°C for 3 hours. The crystals were filtered and displacement washed twice with

1:1 isopropanol/water (25 ml). The product dried with the passage of nitrogen through the cake at 25°C.

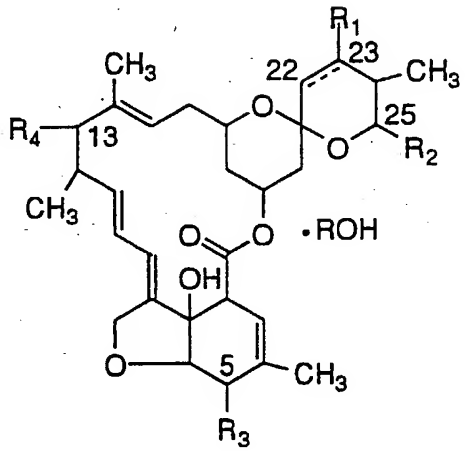
The structure of the 13-O-MEM AVM B1 isopropanol solvate of the present invention is shown below:



The compound crystallizes as an isopropanol solvate in a crystallographic space group $P2_12_12_1$. These compounds show significant long term stability when stored at temperatures ranging from about -10°C to 30°C, preferably from about -5°C to 20°C.

WHAT IS CLAIMED IS:

1. An avermectin C1 to C4 alcohol solvate compound having the formula:



where the broken line indicates a single or a double bond at the 22,23-positions;

R is alkyl of from 1 to 4 carbon atoms resulting in an alcohol consisting of methanol, ethanol, isobutanol, isopropanol, propanol or butanol;

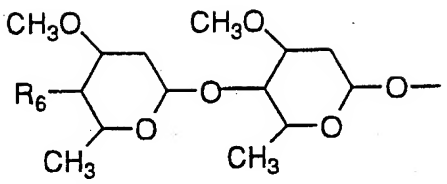
R1 is hydrogen or hydroxy and is hydroxy only when the broken line indicates a single bond;

R2 is alkyl of from 1 to 6 carbon atoms or alkenyl of from 3 to 6 carbon atoms or cycloalkyl of from 3 to 6 carbon atoms;

R3 is hydroxy, methoxy or =NOR5;

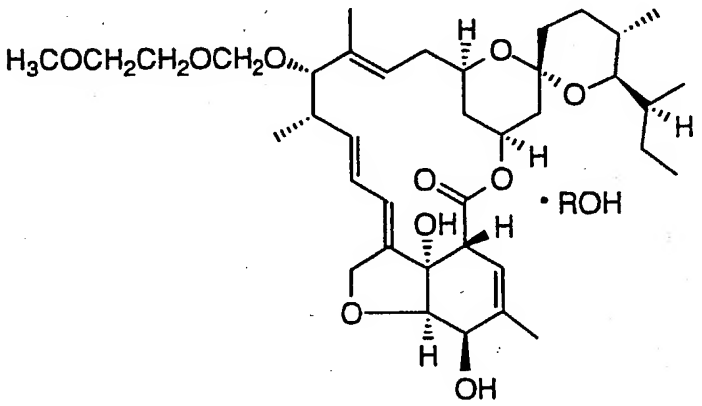
R5 is hydrogen or methyl; and

R4 is hydrogen, (C_1-C_3 alkoxy)(-C0-C_3 alkoxy)methoxy or



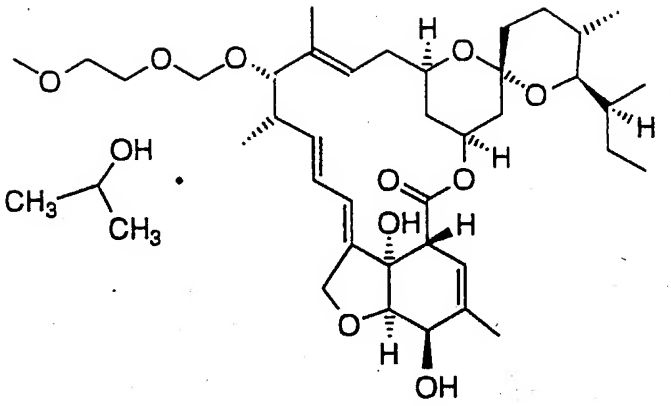
where R_6 is hydroxy, C_1-C_3 amino or C_2-C_3 alkanoylamino.

2. The compound of Claim 1 wherein the alcohol is isopropanol and the avermectin has the formula:

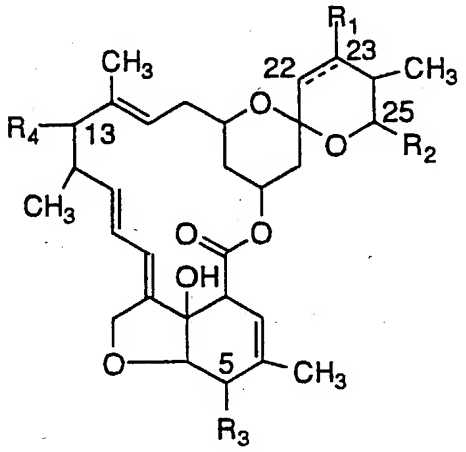


3. A 13-O-MEM AVM B1 isopropanol solvate wherein the crystallographic environment is $P2_12_12_1$.

4. A 13-O-MEM AVM B1 isopropanol solvate compound having the formula:



5. A process for making avermectin alcohol solvates of Claim 1 comprising dissolving one part avermectin having the formula:



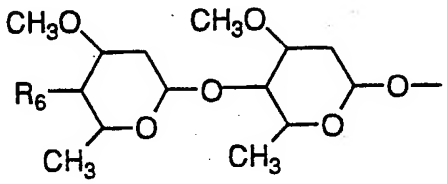
where the broken line indicates a single or a double bond at the 22,23-positions;

R_1 is hydrogen or hydroxy and is hydroxy only when the broken line indicates a single bond;

R_2 is alkyl of from 1 to 6 carbon atoms or alkenyl of from 3 to 6 carbon atoms or cycloalkyl of from 3 to 6 carbon atoms;

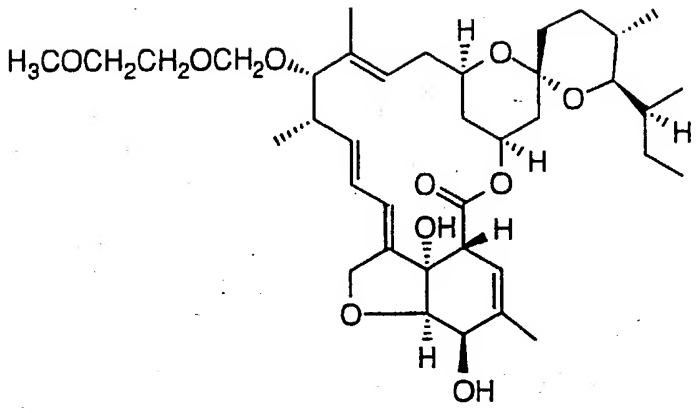
R3 is hydroxy, methoxy or =NOR5;
R5 is hydrogen, or methyl; and
R4 is hydrogen, (C1-C3 alkoxy)(-C0-C3 alkoxy)methoxy

or



where R6 is hydroxy, C1-C3 amino or C2-C3 alkanoylamino, in 1 to 20 parts of an alcoholic solvent selected from the group consisting of methanol, ethanol, butanol, isobutanol, propanol and isopropanol under nitrogen at a temperature from about 50°C to 75°C to form a clear solution, adding to the solution 0 to 20 parts of water while maintaining the temperature above 50°C, cooling the solution to from about 30°C to 40°C, seeding the solution with from about 0.005 grams to 0.1 grams of the avermectin or an alcohol solvate thereof to form a mixture, cooling the mixture to from about 25°C to 15°C and aging the mixture at 25°C to 15°C from about 0 to 24 hours, thereby forming a slurry containing crystals, cooling the slurry to from about 0 to 5°C, aging the slurry at from about 0 to 10°C for about 0 to 6 hours, filtering and washing the crystals with a 1:1 alcohol/water solution to form a wet cake of crystals and drying the crystals under nitrogen at room temperature.

6. The process according to Claim 5 wherein the avermectin compound has the formula:



and the alcoholic solvent is isopropanol.

7. The process according to Claim 5 wherein the avermectin is dissolved in the isopropanol at a temperature from about 60°C to 70°C, and the solution is cooled to from about 35°C to 40°C over a period from about 1/2 hour to 1 1/2 hours.

8. A method for the treatment and prevention of internal and external parasites of animals, which comprises administering the compounds of Claim 1 to an animal.

9. A composition useful for treating animals infected with internal and external parasites which comprises the avermectin compounds of Claim 1 and isopropanol.

INTERNATIONAL SEARCH REPORT

International Application No
PCT/US 94/11247

A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 C07H19/01 A01N43/90 C07D493/22 A61K31/365

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 6 C07H C07D A01N A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	EP,A,0 001 688 (MERCK AND CO., INC.) 2 May 1979 see abstract	1,8,9
A	EP,A,0 001 689 (MERCK AND CO., INC.) 2 May 1979 see abstract & US,A,4 199 569 cited in the application	1,8,9
A	EP,A,0 216 731 (CIBA-GEIGY AG) 1 April 1987 see page 2, line 21 - line 22	1,8,9

☐ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
16 January 1995	25. 01. 95

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Scott, J

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

[redacted] application No.
PCT/US94/11247

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.: 8
because they relate to subject matter not required to be searched by this Authority, namely:

Remark: Although claim 8 is directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

2. [ ] Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. [ ] As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. [ ] As all searchable claims could be searches without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

Information on patent family members

Inte[illegible]nal Application No
PCT/US 94/11247

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
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		AU-A-	4008978	27-03-80
		JP-C-	1443778	08-06-88
		JP-A-	54061197	17-05-79
		JP-B-	62054113	13-11-87
		OA-A-	6063	30-06-81
		US-A-	4206205	03-06-80
		US-A-	4201861	06-05-80
EP-A-0001689	02-05-79	AT-B-	366690	26-04-82
		AU-B-	519569	10-12-81
		AU-A-	3999678	27-03-80
		JP-C-	1668231	29-05-92
		JP-B-	3017837	11-03-91
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		US-A-	4987072	22-01-91